

SEASONALITY OF *Procamallanus (Spirocamallanus) inopinatus* (NEMATODA: CAMALLANIDAE) INFECTION IN *Bryconops melanurus* (CHARACIFORMES: IGUANODECTIDAE)

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ABSTRACT

This study evaluates the host-parasite-environment relationship between the parasitic nematode *Procamallanus (Spirocamallanus) inopinatus* and the fish host *Bryconops melanurus*, as well as reports the seasonal occurrence of the parasite in the host. The study was performed in a total of 78 fish collected from a tributary of Caeté River, Bragança, PA, Northern Brazil. The nematode was present in several organs (stomach, pyloric caecum, anterior intestine, posterior intestine) from 76 out of 78 fish specimens. For each location the prevalence and mean intensity of the infection are provided. The level of parasitism was not influenced by the month of capture, but higher parasitic loads were observed in pyloric caecum and posterior intestine during the rainy season (March to June 2007). Seasonal fish host-nematode relationship can be influenced by fish feeding behaviour patterns associated with flood period variations.

Key words: Amazon; fish; camallanid; parasitology; flooding; host-parasite relationship.

SAZONALIDADE DA INFECÇÃO POR *Procamallanus (Spirocamallanus) inopinatus* (NEMATODA: CAMALLANIDAE) EM *Bryconops melanurus* (CHARACIFORMES: IGUANODECTIDAE)

RESUMO

Este estudo avalia a relação hospedeiro-parasita-ambiente entre o nematoide parasita *Procamallanus (Spirocamallanus) inopinatus* e o peixe hospedeiro *Bryconops melanurus*, bem como relata a ocorrência sazonal do parasita no hospedeiro. O estudo foi realizado em um total de 78 peixes coletados de um tributário do rio Caeté, Bragança, PA, no norte do Brasil. O nematóide estava presente em vários órgãos (estômago, ceco do pilórico, intestino anterior e intestino posterior) de 76 dos 78 espécimes de peixes. Para cada local, a prevalência e a intensidade média da infecção são fornecidas. O nível de parasitismo não foi influenciado pelo mês de captura, mas foram observadas cargas parasitárias mais elevadas no ceco pilórico e no intestino posterior durante a estação chuvosa (março a junho de 2007). A relação sazonal entre o hospedeiro e o nematoide pode ser influenciada por padrões de comportamento alimentar associado às variações do período de inundação.

Palavras-chave: Amazônia; peixes; camalanídeos; parasitologia; inundações; relação parasita-hospedeiro.

INTRODUCTION

Nematodes are frequent parasites of freshwater and marine fish. Usually they inhabit the digestive tract and have low pathogenic importance, but high infections can affect growth and cause mortality (EIRAS, 1994; MARTINS *et al.*, 2004).

Procamallanus spp. were observed in several Brazilian freshwater fishes, with a wide geographical distribution, like *Colossoma macropomum* on Amazon basin (FISCHER *et al.*, 2003), *Cichla piquiti* at Paraná River (FRANCESCHINI *et al.*, 2013), *Serrasalmus marginatus* and *Pygocentrus nattereri* at Pantanal (VICENTIN *et al.*, 2013), and *Hopleryttrinus unitaeniatus*, *Hoplias malabaricus* and *P. nattereri* at Marajó Island

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(BENIGNO *et al.*, 2012) and *Astyanax altiparanae* at São Paulo (ABDALLAH *et al.*, 2012).

Bryconops melanurus Bloch, 1974 is a tropical freshwater fish which has elongated body with upwards mouth facilitating capture of food from the water surface, mainly insects (GAMA and HALBOTH, 2003). Information about the parasitic fauna and host-parasite interactions of *Bryconops* spp. are very scarce. CURRAN (2008) registered two species of the digenean *Creptotrema* in *B. melanurus* from the Aquarico river, Ecuador, and MORAVEC and THATCHER (1997) reported the nematode *Procamallanus (Denticamallanus) dentatus* in *B. alburnoides* from the river Urubu, Amazonia, Brazil. As far as the authors are aware no other studies on the parasitic fauna of *B. melanurus* exist, and an ecological approach of the infection was never performed.

In this study we report an evaluation of the host-parasite-environment relationship and the seasonal distribution of *Procamallanus (Spirocamallanus) inopinatus* (TRAVASSOS *et al.*, 1928), infecting *Bryconops melanurus* in Brazil.

METHODS

A total of 78 fish (2.85 ± 1.02 g mean weight, 6.4 ± 1.1 cm in total length and 5.4 ± 0.8 cm in standard length) were captured (License SISBIO number: 19515) at Chumucuí River, a tributary of Caeté River, at the municipality of Bragança, Pará State, Brazil ($01^{\circ}12'38.3''$ S, $46^{\circ}47'31.7''$ W). The environmental parameters pH (pHmeter Q-400 BC/BD Quimis, Diadema, SP, Brazil), dissolved oxygen, and water temperature (oximeter DO-5510, Lutron, Taipei, Taiwan), were measured at 40 cm depth. Precipitation index was obtained from INMET (National Institute of Meteorology) and the seasons were determined according to SCHAEFFER-NOVELLI and CINTRÓN (1986).

After capture, fish were transported in plastic bag with 1/3 of water and 2/3 of air, in the density of 1 fish L^{-1} to Laboratory from Federal University of Pará, anesthetized by immersion in clove oil solution ($100 \text{ mg } L^{-1}$) and sacrificed by spinal section for biometry and parasitological analysis (EIRAS *et al.*, 2006). The parasites were collected and fixed in AFA solution, quantified (FUJIMOTO *et al.*, 2006), cleared with lactophenol and identified according to TRAVASSOS *et al.* (1928), MORAVEC (1998) and THATCHER (2006). Parasitological indexes were calculated according to BUSH *et al.* (1997): abundance (total number of parasites / total number of examined hosts); (mean intensity = number of parasites / total number of infected hosts) and prevalence ($P\% = \text{total number of infected hosts} / \text{total number of hosts}$) for each organ examined (stomach, pyloric caecum, anterior, medium and posterior intestine) in months. The normality of data was analysed by test D'Agostino and transformed in square root. The t-test ($p=0.01$) for related samples was employed to evaluate the existence of seasonal differences in prevalence and mean intensity values. Pearson's correlations ($p=0.05$) were used to ascertain whether parasitism was influenced by abiotic and biotic parameters, using the Bio Estat 4.0 software.

RESULTS

The dry season occurred between August 2006 to January 2007 and the rainy season between February to July, 2007 (Figure 1a). The water temperature varied from $26.1^{\circ}C$ to $27.5^{\circ}C$ along the year (Figure 1b). The values of pH in dry period (Figure 1c) were alkaline (higher 7.5) and slightly acid in rainy period (6.0-6.9). The highest ($7.15 \text{ mg } L^{-1}$) and lowest values ($3.9 \text{ mg } L^{-1}$) of oxygen concentration were found in November, 2006 and December, 2006 respectively (Figure 1d).

A total of 76 fish were infected with *P. (Spirocamallanus) inopinatus*. Within the host's stomach the highest prevalence (50%) and mean intensity value of infection (5.0 ± 1.5) was found in November, 2006 and June, 2007 respectively. In contrast, to this organ, the lowest prevalence (5.8%) and mean intensity value of infection (1.0 ± 0.7) were found in June, 2007 and November,

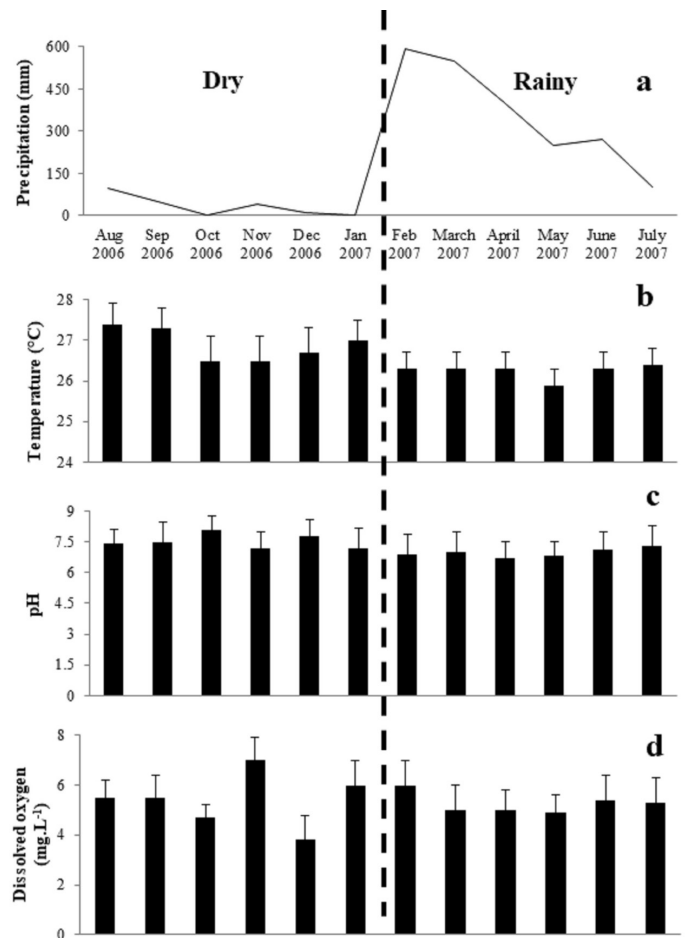


Figure 1. Mean values of rainfall (in mm) in the municipality of Bragança, Pará State, Brazil. (a) Precipitation; (b) water temperature ($^{\circ}C$); (c) pH; and (d) dissolved oxygen ($\text{mg } L^{-1}$), of Chumucuí river – Brazil, between August 2006 and July 2007. Rainfall. Source: Brazilian National Institute of Meteorology (INMET 2007).

2006 respectively (Figure 2a and 2b). No correlation between prevalence and mean intensity was observed.

Intestine was the most parasitized organ. The anterior intestine presented a positive correlation ($r^2=0.47$, $p=0.0135$) with prevalence and mean intensity of infection, higher in January, 2007 (90% prevalence and 13.2 ± 25.1 mean intensity) and lower (33.3% prevalence and 1 ± 0.5 mean intensity) in August, 2006 (Figure 2c and 2d). In contrast, no correlation was found between posterior intestine values and months of sampling.

Posterior intestine showed the highest prevalence (82.3%) and mean intensity (5.8 ± 5.1) in June, 2007, with lower values of prevalence (30%) and higher of mean intensity of infection (12.6 ± 8.4) in January, 2007 (Figure 2e and 2f).

Parasitic infection in pyloric caecum was higher in August, 2006 (33.3% prevalence and mean intensity of 1.0 ± 0.5) followed by a reduction in February, 2007 (prevalence 11.1% and mean intensity 5.0 ± 1.6), as demonstrated in the stomach and posterior intestine with no correlation (Figure 2g and 2h).

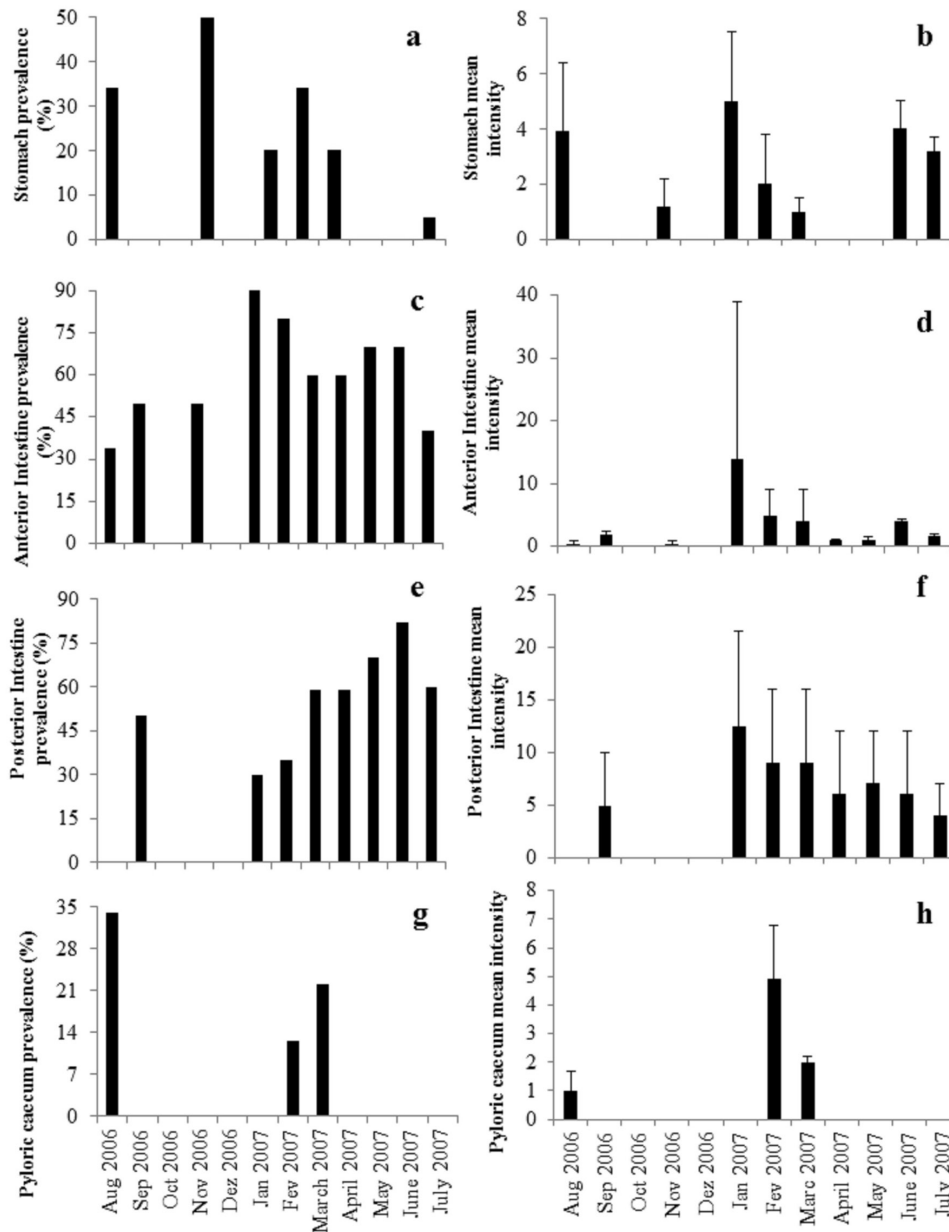


Figure 2. Mean values of prevalence rate (%) and mean intensity of infection by *Procamallanus (Spirocamallanus) inopinatus* in the stomach (a, b), anterior intestine (c, d), posterior intestine (e, f) and pyloric caecum (g, h) of *Bryconops melanurus* from Caeté river, Northern Brazil.

In relation to flooding and rainy season the pyloric caecum showed the highest prevalence in the rainy period, which was significantly different from the dry one (Figure 3). Weight and length did not present correlation with prevalence and mean intensity of infection (Table 1).

Values of prevalence in the stomach was positively correlated to oxygen concentration ($r^2=0.70$) contrarily to that observed in pyloric caecum that showed a negative correlation ($r^2=-0.65$). The mean intensity of infection observed in the posterior intestine presented negative correlation ($r^2=-0.65$) with pH (Table 2). Furthermore, positive correlation was found between stomach and anterior intestine ($r^2=0.62$), as well as pyloric caecum and anterior intestine ($r^2=0.54$) (Table 3).

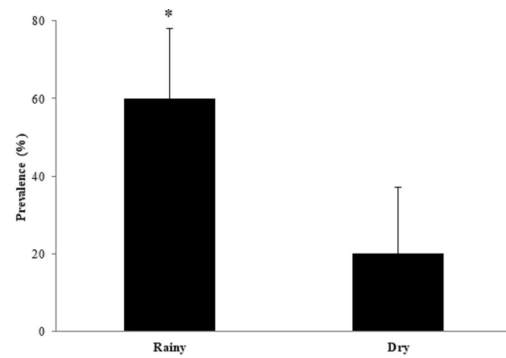


Figure 3. Prevalence values of *Procamallanus (Spirocamallanus) inopinatus* in the pyloric caecum in the rainy and dry period between August 2006 and July 2007. *Significant difference ($p < 0.01$).

Table 1. Pearson's correlation among weight, total length, standard length and mean intensity of infection of *Procamallanus (Spirocamallanus) inopinatus* in *Bryconops melanurus* from Caeté river, Northern Brazil.

Site of infection x weight	Prevalence (r values)			Mean intensity (r values)		
	Weight	Total length	Standard length	Weight	Total length	Standard length
Stomach	0.34	0.29	0.13	-0.33	0.35	-0.35
Anterior intestine	-0.01	-0.33	-0.32	-0.15	0.38	-0.36
Posterior intestine	-0.07	0	0.06	0.16	0	0.05
Pyloric caecum	-0.44	-0.41	-0.30	-0.25	0.49	-0.41

Table 2. Pearson's correlation among the values of pH, dissolved oxygen and water temperature with prevalence rates and mean intensity of *Procamallanus (Spirocamallanus) inopinatus* in *Bryconops melanurus* from Caeté river, Northern Brazil.

Site of infection	Prevalence (%)		
	pH	Oxygen	Temperature
Stomach	0.22	0.70*	0.03
Anterior intestine	-0.03	-0.21	-0.09
Posterior intestine	-0.23	-0.14	0.27
Pyloric caecum	-0.24	-0.65*	-0.43
Site of infection	Mean intensity		
	pH	Oxygen	Temperature
Stomach	0.48	0.20	0.36
Anterior intestine	0.15	-0.05	0.33
Posterior intestine	-0.65*	-0.09	-0.19
Pyloric caecum	-0.26	-0.50	0.02

*Significant difference ($p < 0.05$).

Table 3. Pearson's correlation among *Procamallanus (Spirocamallanus) inopinatus* infection on different organs of *Bryconops melanurus* from Caeté river, Northern Brazil.

Site of infection	Stomach	Anterior intestine	Posterior intestine	Pyloric caecum
Stomach	10.000	-	-	-
Anterior intestine	0.6282*	10.000	-	-
Posterior intestine	0.3685	0.0023	10.000	-
Pyloric caecum	0.2386	0.5468*	-0.0026	10.000

*Significant difference ($p < 0.05$).

DISCUSSION

In rainy season, there is a great increase of the flooded surface providing a large amount of organic matter to the ecosystem and influencing the biological cycle of animals (LIZAMA *et al.*, 2006; APRILE and DARWICH, 2013) including the life-cycle of fish parasites. In the studied area, there was a clear delineation between the highest and lowest rainfall seasons that may have interfere with the rivers dynamic and the characteristics of the parasitic fauna of *B. melanurus*.

Bryconops melanurus feed mainly on terrestrial insects that fall from riparian vegetation (PEREIRA *et al.*, 2007). Once *P. (Spirocamallanus) inopinatus* has not a life cycle involving terrestrial insects it is supposed that other preys, like copepods, which may be intermediate hosts for the parasite may be included in the diet of the fish (SILVA *et al.*, 2008). Copepods are recognized as intermediate host for *Procamallanus* spp. (MORAVEC, 1994; DE and MAITY, 2000), and this supports the idea that the higher prevalence of *P. (S.) inopinatus* in the rainy season can be associated with feeding on intermediate hosts of the parasite.

Another tentative explanation for the infection would be the zooplanktonic habits in the younger stages of fish life. However, this is not probable once no correlation was observed between parasite load, size, and weight of fish, and a seasonal accumulation of the parasites due to the age and size as suggested by SABAS and LUQUE (2003) for other fish species was not observed in the present case.

The production dynamics of Chumucuí River is higher in periods of high precipitation (GORAYEB *et al.*, 2010). There is a relationship between physicochemical characteristics of water and variation of intensity of parasitism, due to the increase of aquatic productivity (GOATER *et al.*, 2005). The environmental modifications related to rainfall, namely the variation in the amount of dissolved oxygen, as well as changes of the water pH resulting from the decomposition of tonnes of leaves may indirectly be related with fish parasitic fauna.

In what concerns the positive correlation observed among different organs, it is probable that the infection in stomach and the pyloric caecum influenced the infection in anterior intestine or vice versa. Thus, infection in the pyloric caecum during the rainy season could be caused by parasites distribution along the intestinal tract, and no exactly through direct effect of environmental conditions.

Thereby, due to higher number of parasites present in the anterior intestine (main infection site), it is possible that infection on this site influenced the infection in the stomach and pyloric caecum by migration of parasites. A similar pattern was also verified in *Macrodon ancylodon* (FUJIMOTO *et al.*, 2012). POULIN (2001) described this relocation of the infection by migration due to interspecific competition where the most numerous species compete for the same niche leading to minor species to search for new places of infection with less competition, which was called as “niche realized”. Similarly to the interspecific competition, an intraspecific competition could force the present specimens to extend their distribution throughout the digestive tract. This report was the first record of *P. (S.) inopinatus* in *B. melanurus* therefore more studies are needed to elucidate this question and the relative importance of the different mechanisms involved on the parasites location.

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